

# Chronic Hæmolysis Occurring in Patients Following Cardiac Surgery

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Since 1961 a number of reports have been published in which significant hæmolysis has been described in patients following cardiac surgery with extracorporeal circulation. In the majority of cases prosthetic valves had been inserted. In this report we present results of a study made to determine the frequency of minimal as well as overt hæmolysis in a series of patients who underwent open-heart surgery for correction of a variety of defects. It appeared that mild chronic hæmolysis was a consistent finding in patients in whom a prosthetic valve was inserted. In addition, hæmolysis was occasionally demonstrated in patients with other intracardiac deformities.

## SUBJECTS AND METHODS

The 39 patients had cardiac surgery performed with the use of cardiopulmonary bypass between February 1961 and December 1963. Of these, 27 had valve replacements, 25 with Starr-Edwards valves and 2 with dacron cusps. Starr-Edwards valves were inserted in 13 patients to replace the mitral valve; in 11 patients the aortic valve and in one patient both mitral and aortic valves were replaced. Dacron cusps were used in both instances to replace damaged aortic valves.

Of the remaining 12 patients, 6 had had surgical repair of an atrial septal defect with use of a silk suture, 2 had had surgical repair of a ventricular septal defect, one with a silk suture and a teflon patch and the other with a dacron patch. The other cases included one each of correction of rheumatic mitral insufficiency by means of mitral annuloplasty, one mitral leaflet advancement using ivalon pledgets, one surgical correction of a tetralogy of Fallot, and one repair of a rupture of the sinus of Valsalva.

The patients ranged in age from 10 to 58 years and were about evenly divided between the two sexes. There were 19 patients who had been operated on before July 1963 and who were not available for detailed

study. The investigation of these was limited, therefore, to examinations that could be done on mailed blood specimens, namely, measurement of serum haptoglobin, Schumm's test for methæmalbumin, and examination of a blood smear. This initial study suggested that chronic hæmolysis was occurring in our patients with Starr-Edwards valvular prostheses or dacron cusps, and this prompted a more detailed study on 20 patients operated on in the last four months of 1963. In this latter group, pre-operative estimations of serum haptoglobin and serial determinations post-operatively of serum haptoglobin and methæmalbumin, urine hæmosiderin, reticulocyte count, and examination of blood smears were carried out. All patients were followed for at least 14 days except for four in whom the serum haptoglobin had returned to 100 mg./100 ml. or greater within the two weeks after operation. Only one patient was suspected on clinical grounds of having a hæmolytic anæmia.

Blood films were made directly from blood obtained from the antecubital vein or by fingerprick. They were stained with May-Gruenwald-Giemsa or Wright's stain and carefully examined for presence of distorted or fragmented erythrocytes. The numbers of distorted or fragmented erythrocytes were assessed using a modification of the method of Zipursky *et al.* (1963).

Serum haptoglobin levels were determined using the method of Nyman (1959), as modified by Javid and Horowitz (1960). The normal range is 60 to 160 mg./100 ml. plasma.

Haptoglobins are a group of proteins present in the plasma, which bind free hæmogoblin, and their level in plasma is regarded as a sensitive index of intravascular hæmolysis (Dacie, 1960; Jayle and Moretti, 1962). The amount of hæmoglobin that can be bound by the haptoglobins in normal subjects has been estimated to be in the range of 60 to 160 mg./100 ml. plasma. When this level is exceeded, free hæmoglobin appears in the urine. Once the hæmoglobin-haptoglobin complex has been formed, it is cleared from the plasma in a few hours and the haptoglobin which is lost is only slowly regenerated, taking about a week to return to normal values in the absence of any further hæmolysis (Jayle and Moretti,

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1962). In the presence of chronic hæmolytic, the haptoglobin is continually cleared from the plasma with resulting low or zero levels (Dacie, 1960). Raised haptoglobin levels, thought to result from increased production, are found in association with a variety of inflammatory or neoplastic diseases (Jayle and Moretti, 1962). A Schumm's test was also done on all the specimens to determine possible presence of methæmalbumin, a pigment not normally present in plasma except following intravascular hæmolytic (Dacie, 1960). It appears when the amount of free hæmoglobin entering the plasma exceeds the binding capacity of haptoglobin. The presence of this latter pigment implies, therefore, a somewhat greater degree of hæmolytic than is indicated by a reduced level of haptoglobin alone.

TABLE  
ESTIMATIONS OF HAPTOGLOBIN IN PATIENTS WITH  
VALVULAR PROSTHESES AND AFTER OTHER  
OPERATIONS

Values of haptoglobin (mg./100 ml.)	Valve replacement		Other cardiac operations	
	Pre-op. (11)	Post-op. (27)	Pre-op. (9)	Post-op. (12)
0	2	20	—	2
< 60	4	6	3	—
> 60	5	1	6	10
Abnormal RBC morph.	—	8	—	3
Reticulocytes > 2 per cent	—	71/0	—	2

### RESULTS

The results of the investigation are shown in the Table. In the 25 patients with Starr-Edwards valvular prostheses, the most recent measurement of the serum haptoglobins showed them to be absent in 18 and less than 60 mg./100 ml. in six. One patient had a normal amount of haptoglobin. Two patients who had aortic dacron cusps also had no measureable haptoglobin. In contrast, of 12 patients on whom other types of cardiac surgery had been performed, 10 had normal or above normal levels of serum haptoglobins and in only two patients were these proteins not found. The measurements were made over a period of 14 to 814 days after operation.

Pre-operative values of the serum haptoglobins were determined on 11 patients who later had Starr-Edwards valves inserted, and on 9 patients subjected to other types of cardiac surgery. In the patients in whom artificial valves were later inserted, the serum haptoglobins were absent in 2, less than 60 in 4, normal in 4, and raised in one. In the 9 patients who had other cardiac operations and on whom haptoglobins were measured before operation, values were normal in 6 and reduced in 3.

In the Figure the results of the serum haptoglobin analyses have been plotted. Approximately half of

the 20 values determined before operation were below 60 mg./100 ml. There appeared to be no correlation between the various cardiac lesions and the level of serum haptoglobin. After operation, there appeared to be a considerable variation in the levels of this protein. In the first 24 hours the haptoglobin was usually greatly reduced, probably as a result of hæmolytic occurring during the cardiopulmonary bypass. By the second day, the values for several patients in whom artificial cardiac valves were not inserted had returned to normal and, with the exception of 2 patients, the levels beyond the fourth day were normal or raised. One of the two patients without valvular prosthesis in whom low haptoglobin levels persisted was a 33-year-old woman who had a repair of an atrial septal defect secundum, with silk suture. The other was a 58-year-old man who had a mitral advancement with the use of ivalon pledgets.

In one patient with a valvular prosthesis, a level of 100 mg. haptoglobin was found 17 days after operation. A repeat determination done on the 59th day showed haptoglobin to be absent.

Three or more urine specimens were examined in 17 patients to detect the presence of hæmosiderin. Single specimens of urine were examined in an additional 9 patients. Hæmosiderin in the urine is regarded as an indication of intravascular hæmolytic (Crosby and Dameshek, 1951; Dacie, 1960). Moderate hæmosiderinuria occurred in the one patient in whom hæmolytic anæmia was suspected clinically. Trace amounts of hæmosiderin were found in a few urine specimens of patients with and without valvular prostheses.

In the patients in whom reticulocyte counts were obtained 14 or more days after operation, only 7 showed values above normal. The highest count, 4.1 per cent, was found in the patient who had clinical evidence of a hæmolytic anæmia. This patient was the only one of our patients who had a combined aortic and mitral valve replacement. The presence of a hæmolytic anæmia was suggested by a persistent anæmia without evidence for blood loss, which necessitated transfusions totalling 3000 ml. blood in the first six weeks after operation. No haptoglobin was found in the serum, and methæmalbumin was present. This patient also had moderate hæmosiderinuria and slight distortion and fragmentation of his erythrocytes. In addition, he developed lymphocytosis with atypical lymphocytes, mild pyrexia, and splenomegaly 120 days after operation. This was diagnosed as post-cardiotomy lymphocytosis (Wheeler, Turner, and Scannell, 1962). However, it seemed unlikely that this was related to the hæmolytic. Further studies, 217 days after his operation, showed absent serum

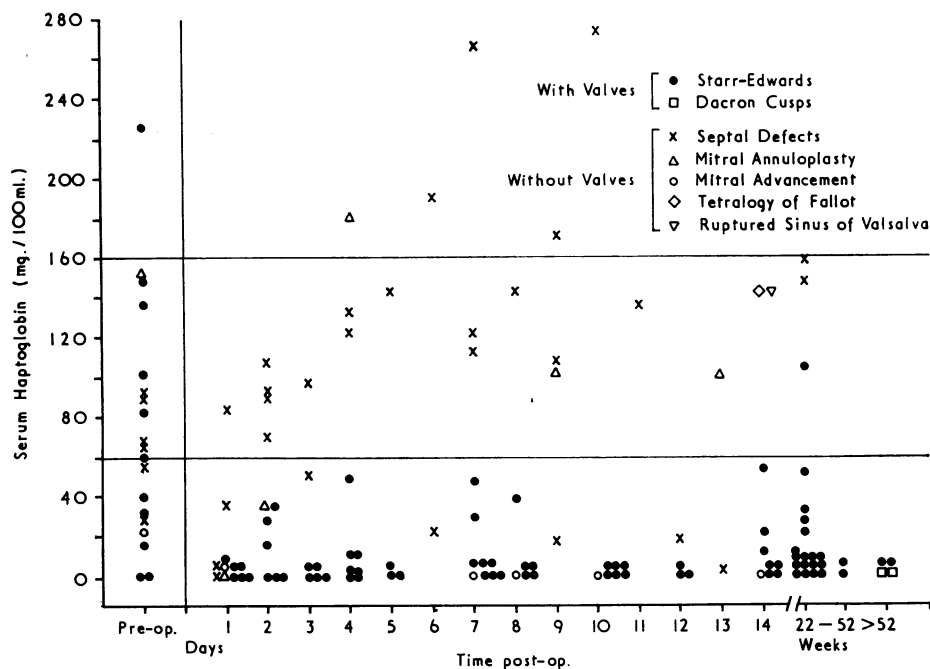


FIG.—Serum haptoglobin values before and after operation on patients after open-heart surgery, showing low values in patients with valvular prostheses as compared with patients without prostheses.

haptoglobins, no methæmalbuminæmia, only slight hæmosiderinuria, and a reticulocyte count of 1.8 per cent. By then he was not anæmic and no longer required blood transfusion. This patient died after a second operation but permission for necropsy was not obtained.

Blood films for all patients were examined and only doubtful evidence of erythrocyte distortion and fragmentation of an abnormal degree was found in the majority, but in two patients there was a considerable increase in the numbers of these abnormal erythrocytes. One of these had a valvular prosthesis, and in this patient haptoglobin was not demonstrated in the serum. The other, a patient who had a repair of a ventricular septal defect, had a normal haptoglobin.

Methæmalbumin was present in sera of only 9 patients: the amount was not measured. Eight of these patients had Starr-Edwards valves and persistently low or absent haptoglobins; the other patient had a repair of an atrial septal defect. At the time methæmalbumin was detected on the tenth day after operation, the haptoglobin level was raised (308 mg./100 ml.).

#### DISCUSSION

Low or absent haptoglobins in plasma are regarded as an indication of increased red cell destruction (Nosslin and Nyman, 1958; Dacie, 1960).

The lack of haptoglobins in the patients with Starr-Edwards valvular prostheses in contrast to patients who had other types of surgical procedures on the heart suggests that there is a constant mild hæmolysis of erythrocytes in the patients with these valves. Additional evidence for this is provided by the presence of methæmalbumin in the serum and the hæmosiderinuria observed in some of the patients.

The clinical evidence of hæmolysis in the one patient who had combined aortic and mitral valve replacement gives further support to this suggestion. While we have been able to study only two patients with aortic dacron cusps, the absence of haptoglobins in these patients suggests they have a similar disturbance.

Some degree of hæmolysis is recognized as occurring with the use of cardiopulmonary bypass (Stewart and Sturridge, 1959). While this would account for the lack of haptoglobin and methæmalbuminæmia immediately after operation, it would not account for these findings after many months.

Reerink-Brongers, Prins, and Krijnen (1962) found low levels of haptoglobin when the  $^{51}\text{Cr}$  half-time was less than 20 days ( $T_{1/2}^{51}\text{Cr}$ ). This represents a destruction rate of greater than twice normal (Donohue *et al.*, 1955). This was based on studies of patients with hæmolytic anæmia where the cell

destruction was thought to be predominantly extravascular. Nosslin and Nyman (1958) reported an absence of haptoglobin in 34 out of 36 patients in whom the  $T\frac{1}{2}$   $^{51}\text{Cr}$  was less than  $17\frac{1}{2}$  days. Dacie (1960) quoted a study by Brus and Lewis, which showed that haptoglobin level in the serum fell to zero when the hæmoglobin turnover exceeded twice the normal rate. From these studies it appears that the lack of haptoglobins in the patients in this study could represent a significant hæmolysis. The limited reticulocyte counts available in this study tend to support this estimate. Persistent ahaptoglobinæmia with iron deposition in the proximal renal tubules does not adversely affect renal function, and whether or not the ability of haptoglobin to bind hæmoglobin is physiologically significant has not been clearly established (Whitten, 1962; Andersen, Gabrieli, and Zizzi, 1965).

There have been a number of reports of patients who have developed overt hæmolysis following open-heart surgery (Andersen *et al.*, 1965; Brodeur *et al.*, 1965; DeCesare, Rath, and Hufnagel, 1965; Gehrmann and Loogen, 1964; Kezdi, Head, and Buck, 1964; Pirofsky *et al.*, 1965; Reed and Dunn, 1964; Sayed *et al.*, 1961; Sears and Crosby, 1965; Sigler *et al.*, 1963; Stewart and Sturridge, 1959; Verdon, Forrester, and Crosby, 1963; Viner and Frost, 1965). Of 57 patients so far reported, 8 had repairs of defects without insertion of valves and the remainder had one or more valves replaced. Of these, 30 were aortic prostheses, all but 24 of them ball-valve type. Ten had mitral valves, and 3 had both aortic and mitral valves replaced. In one report the valve affected was not stated. Evidence of hæmolysis appeared most commonly in the immediate post-operative period, but in 2 patients, both with dacron cusps, it developed suddenly at 6 and 11 months. The destruction of erythrocytes was sufficient in these cases to produce moderate to severe anæmia, often requiring blood transfusions. Evidence of intravascular hæmolysis in the form of raised hæmoglobin or methæmalbumin in the serum, hæmosiderinuria, and fragmentation and distortion of the erythrocytes, was demonstrated in many of these patients. Serum haptoglobin was recorded as absent or very low in all 29 patients in whom it was estimated.

Brodeur *et al.* (1965) studied the erythrocyte survival in 21 patients with aortic valvular disease without prostheses, 12 with aortic ball valve prostheses alone, and 8 with multiple prostheses. The  $^{51}\text{Cr}$  half-times in all 20 patients with aortic disease without prostheses was reduced, with a mean value of 20.75 days (normal 27—3 days). The mean survival in patients with valvular prostheses, either

single or multiple, was similarly reduced (mean 20 days).

In contrast to such patients in whom overt hæmolysis was present, the patients reported in this study showed evidence of unusual hæmolysis which occurred predominantly in patients with Starr-Edwards valves and much less frequently in patients following other types of cardiac surgery. The differences between our patients and those reported by other workers appears to be one of degree, our findings indicating a subclinical rather than an overt hæmolysis.

There are several possible explanations for the hæmolysis which appears to be taking place in these patients. Sayed *et al.* (1961) proposed that the hæmolysis in their patient resulted from damage to the erythrocytes produced by a jet of blood striking a bare teflon patch. They produced some evidence, based on *in vitro* studies, that teflon had a harmful effect on erythrocytes. Evidence is accumulating to show that erythrocytes are quite susceptible to damage on exposure to foreign surfaces and possibly even to damaged endothelium (Brain, Dacie, and Hourihane, 1962). Starr-Edwards valves are so placed that the teflon ring usually becomes endothelialized within a few days. The metal and the silastic ball remain exposed. The observation that all the patients in whom overt hæmolytic anæmia has occurred either had teflon used in the repair or have had insertion of artificial valves lends further support to the proposal that the foreign material may be a factor contributing to the hæmolysis. One of the two patients in our series without a valvular prostheses and absent haptoglobin had an atrial septal defect secundum repaired with silk suture; the other had ivalon pledgets inserted for mitral advancement, and later developed clinical evidence for breakdown of the repair.

Mechanical damage to the erythrocyte by the action of the ball-valve through direct injury is also a possibility. A somewhat similar type of prosthesis made of lucite has been shown to produce hæmolysis when inserted into the aorta of dogs (Stohlman *et al.*, 1956). Pirofsky *et al.* (1965) have suggested, on the basis of finding positive Coombs tests in 6 of 7 patients with hæmolytic anæmia following the insertion of a Starr-Edwards valve, that the hæmolysis in some cases is a manifestation of an auto-immune reaction initiated by the exposure of antigenic sites on the erythrocyte surface by the action of the ball-valve. These and three patients in another series that they studied (Brodeur *et al.*, 1965) are the only ones in whom positive Coombs tests have been demonstrated.

Sigler *et al.* (1963) suggested that the major cause

of the haemolysis was turbulence in blood flow resulting from altered intracardiac flow. In support of this explanation is our finding of low haptoglobins before operation in a number of patients. These comprised patients who had gross mitral insufficiency, large shunts through atrial septal defects, and patients in whom cardiac repairs had broken down.

Patients with Starr-Edwards valves have murmurs which suggest some turbulence. The presence of a systolic ejection murmur has been a consistent post-operative observation in our patients who have had aortic valves replaced. That some obstruction to flow may occur with higher cardiac output has been suggested by our observation in patients with mitral valve replacement that the wedged pulmonary arterial pressure has risen to abnormal levels upon exercise during late post-operative catheterization studies. Further evidence in this respect has been provided by Sears and Crosby (1965) who showed a consistent relation between the rate of haemolysis and physical activity in two patients who had cardiac surgery.

The evidence for chronic mild haemolysis in a number of our patients before operation, the persistence of low or absent haptoglobin levels in two patients who had no prosthetic valves inserted, and the observation that a pressure gradient may exist across many of the prosthetic valves at higher flow rates (Kezdi *et al.*, 1964) all support the suggestion that turbulence in blood flow is a major factor which leads to haemolysis in these patients. The demonstration of reduced red cell survival in patients who had aortic valvular disease but had not been operated upon also appears to support the concept that turbulence rather than the valve itself may be responsible for the haemolysis.

#### SUMMARY

We have studied 25 patients with cardiac valvular prostheses, 2 with dacron aortic cusps, and 12 patients who had undergone cardiac surgery without insertion of a prosthesis, to determine whether or not the artificial valves, turbulence, or other factors have an adverse effect on the patient's erythrocytes, reducing their life span.

The reports of overt haemolysis occurring after open-heart surgery have been reviewed to compare the findings in these patients with the ones comprising this study.

Using a deficiency of serum haptoglobins as a criterion of excessive erythrocyte destruction, chronic subclinical haemolysis appears to be an almost constant finding in patients with Starr-Edwards valves.

It seems unlikely that haemolysis is attributable only to the presence of foreign material, because haemolysis was also detected in patients studied before operation.

The degree of haemolysis is not known. It may, however, be of a significant amount, possibly increasing the erythrocyte destruction rate twofold.

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